Supplement

Figure S1

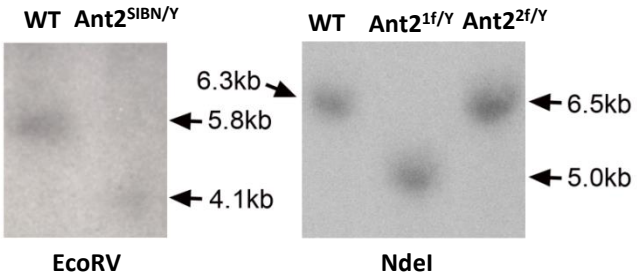


Figure S2

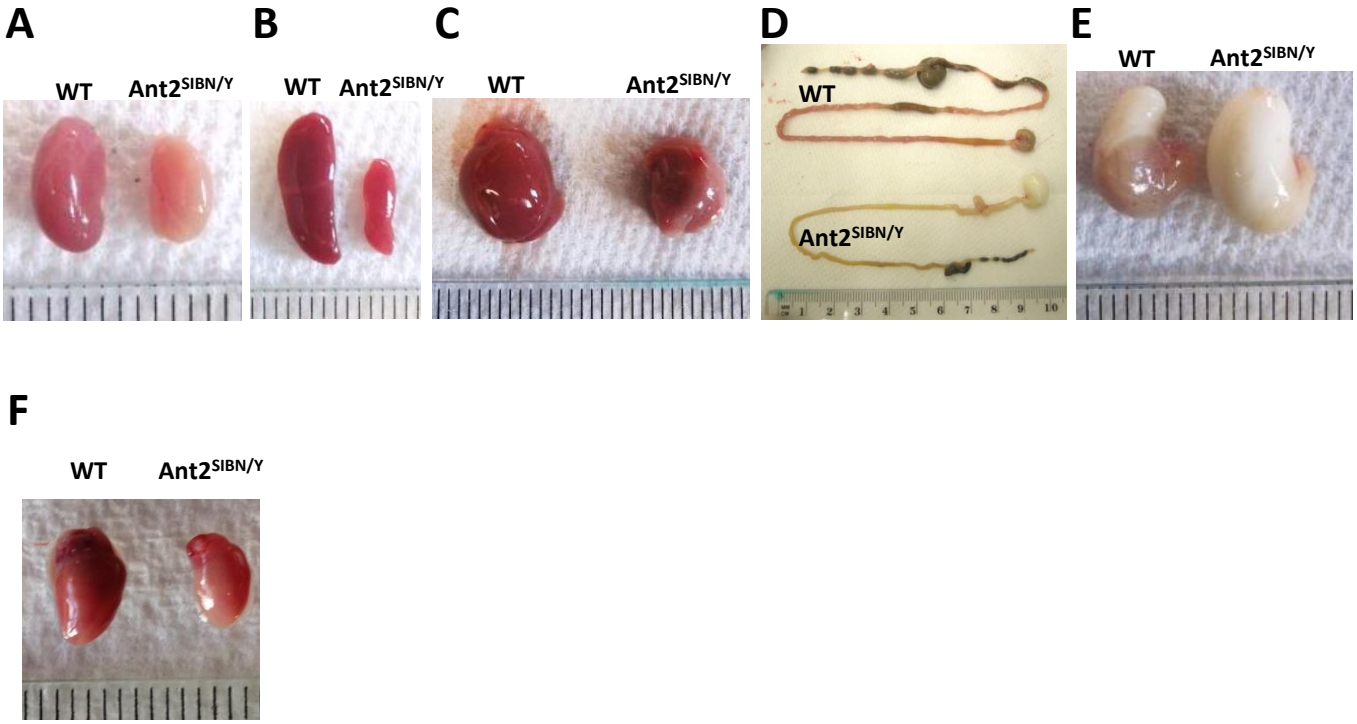

A
WT
Ant2SIBN/Y
B
WT
Ant2SIBN/Y
C
WT
Ant2SIBN/Y
D
WT
Ant2SIBN/Y
E
WT
Ant2SIBN/Y
F
WT
Ant2SIBN/Y

Figure S3

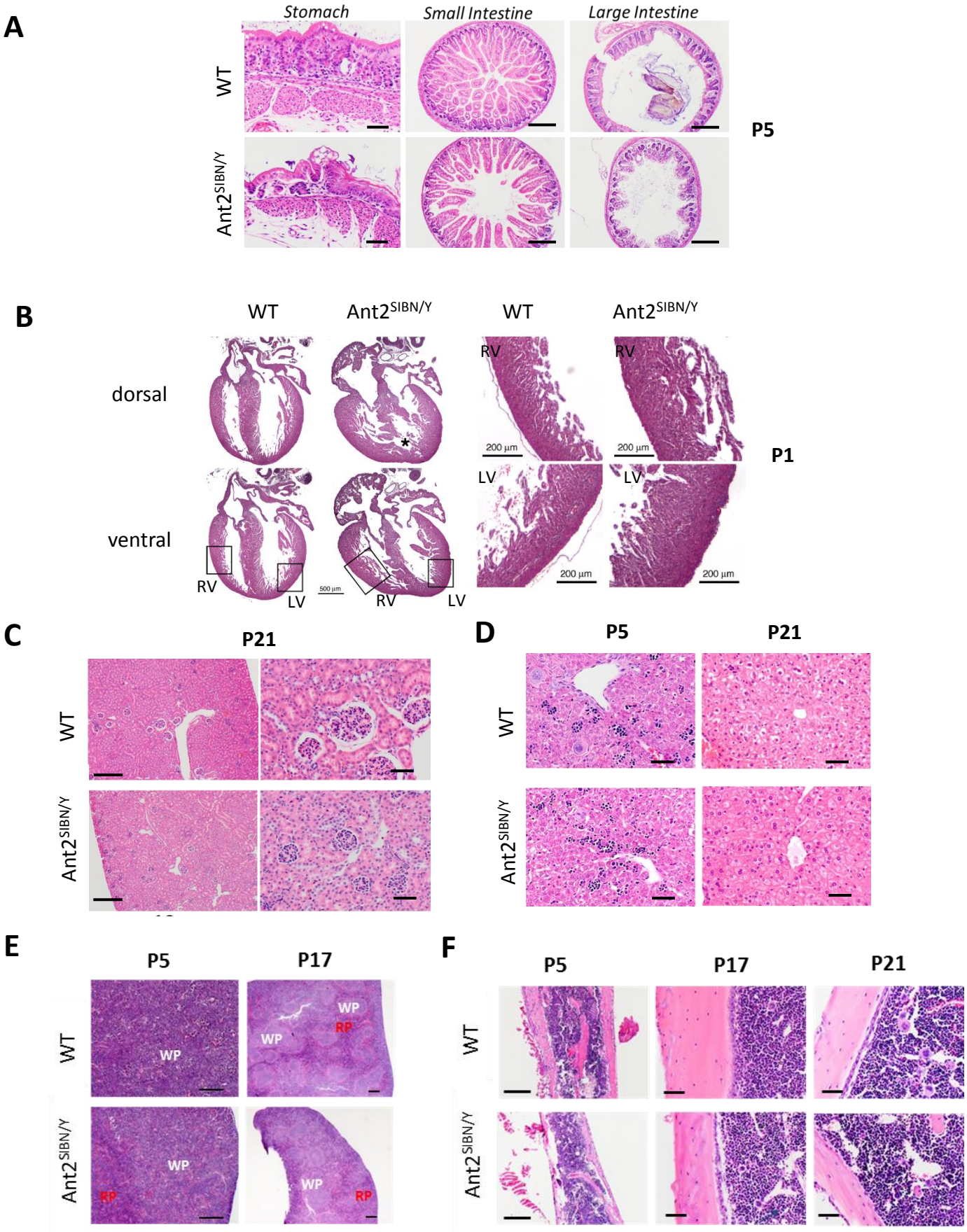

A
Stomach
Small Intestine
Large Intestine
WT
Ant2SIBN/Y
P5
B
WT
Ant2SIBN/Y
WT
Ant2SIBN/Y
dorsal
ventral
RV
LV
500 µm
200 µm
P1
C
P21
WT
Ant2SIBN/Y
D
P5
P21
WT
Ant2SIBN/Y
E
P5
P17
WT
Ant2SIBN/Y
WP
RP
F
P5
P17
P21
WT
Ant2SIBN/Y

Figure S4

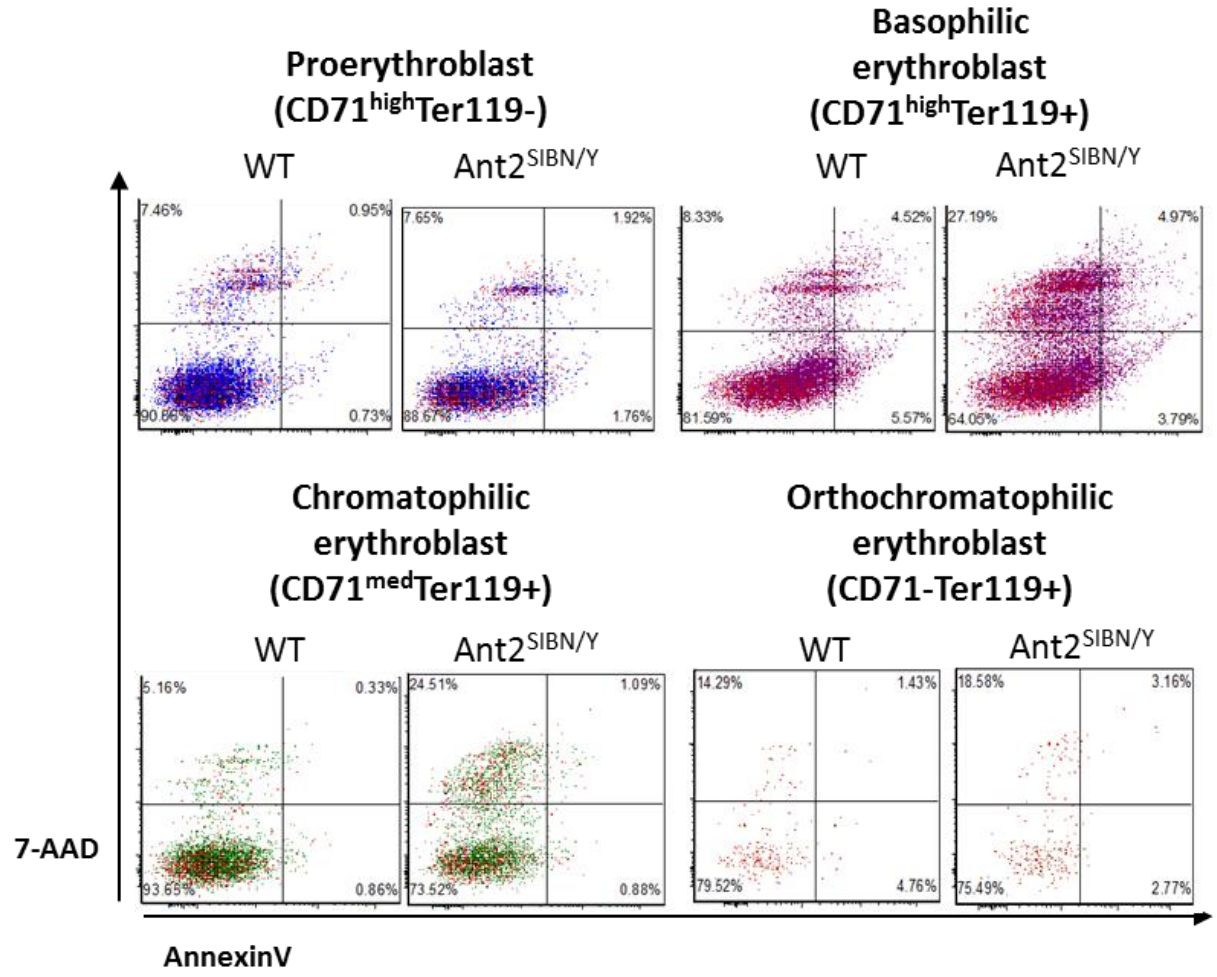

Proerythroblast
(CD71highTer119-)
WT
Ant2SIBN/Y
7.46%
0.95%
90.66%
0.73%
7.65%
1.92%
88.67%
1.76%
Basophilic
erythroblast
(CD71highTer119+)
WT
Ant2SIBN/Y
8.33%
4.52%
81.59%
5.57%
27.19%
4.97%
64.05%
3.79%
Chromatophilic
erythroblast
(CD71medTer119+)
WT
Ant2SIBN/Y
5.16%
0.33%
93.65%
0.86%
24.51%
1.09%
73.52%
0.88%
Orthochromatophilic
erythroblast
(CD71-Ter119+)
WT
Ant2SIBN/Y
14.29%
1.43%
79.52%
4.76%
18.58%
3.16%
75.49%
2.77%
7-AAD
AnnexinV

Figure S5

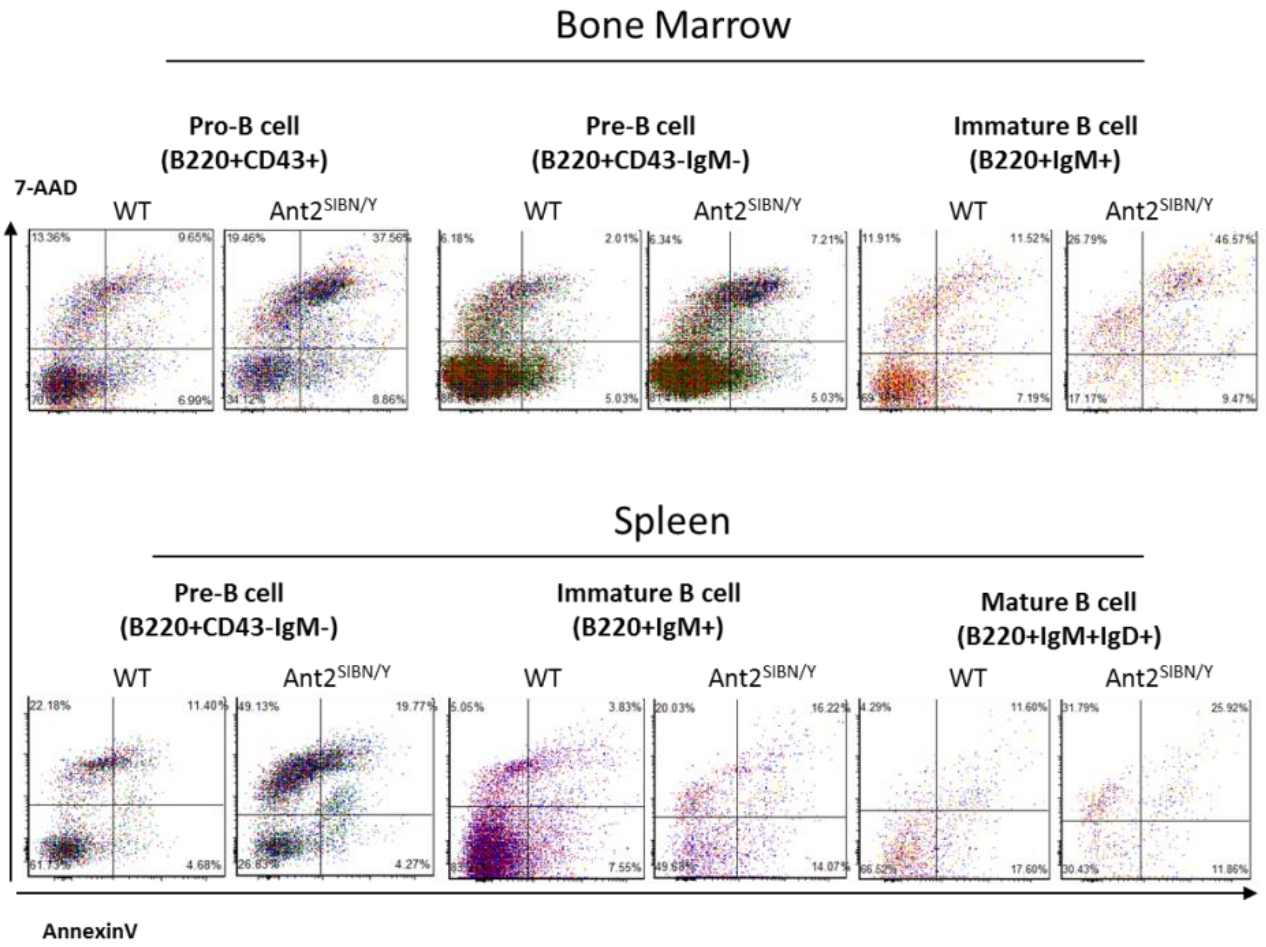

Bone Marrow
Pro-B cell
(B220+CD43+)
Pre-B cell
(B220+CD43-IgM-)
Immature B cell
(B220+IgM+)
7-AAD
WT
Ant2SIBN/Y
13.36%
9.65%
6.99%
19.46%
37.56%
8.86%
6.18%
2.01%
5.03%
6.34%
7.21%
5.03%
11.91%
11.52%
7.19%
26.79%
46.57%
17.17%
9.47%
Spleen
Pre-B cell
(B220+CD43-IgM-)
Immature B cell
(B220+IgM+)
Mature B cell
(B220+IgM+IgD+)
22.18%
11.40%
4.68%
49.13%
19.77%
4.27%
5.05%
3.83%
7.55%
20.03%
16.22%
14.07%
4.29%
11.60%
17.60%
31.79%
25.92%
11.86%
AnnexinV

Figure S6

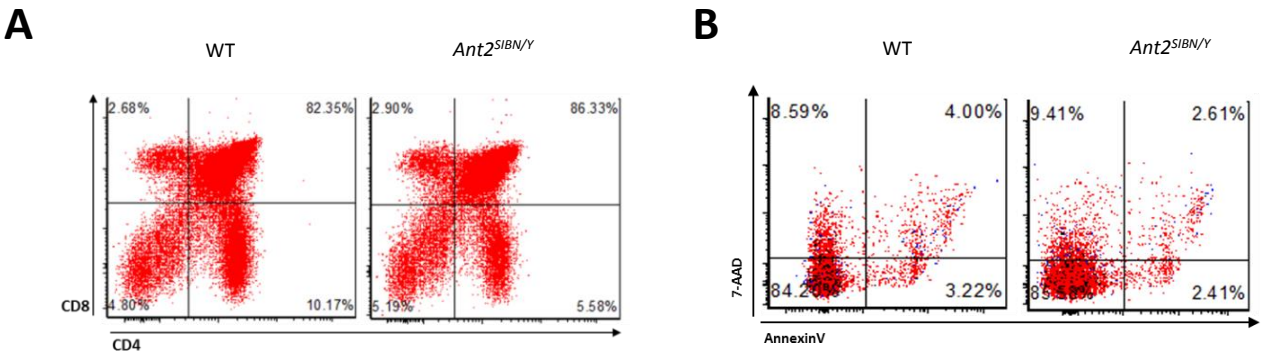
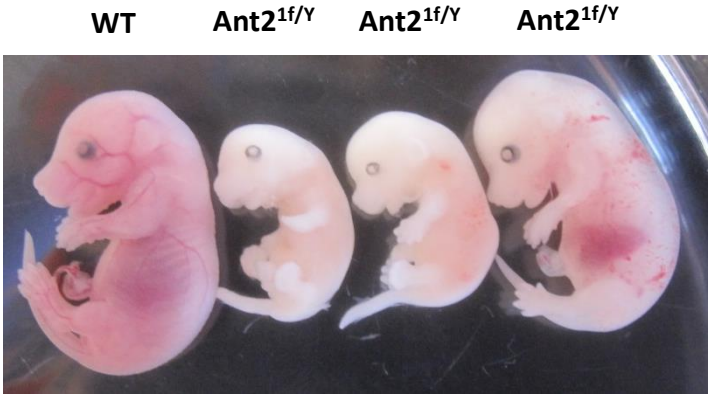

A
WT
Ant2SIBN/Y
2.68%
82.35%
2.90%
86.33%
CD8
4.80%
10.17%
5.19%
5.58%
CD4
B
WT
Ant2SIBN/Y
8.59%
4.00%
9.41%
2.61%
7-AAD
3.22%
2.41%
AnnexinV

Figure S7



SUPPLEMENTAL FIGURE LEGEND

Figure S1: Genotyping of the targeted *Ant2* alleles by Southern blot analysis.

Genomic DNA was extracted from *Ant2*$^{+/Y}$ (WT), *Ant2*$^{SIBN/Y}$, *Ant2*$^{2f/Y}$ and *Ant2*$^{1f/Y}$ ES cells, digested with the indicated restriction enzymes, and blotted with the probe indicated in the Figure1A.

Figure S2: Macroscopic images of isolated organs from representative WT and *Ant2*$^{SIBN/Y}$ mice.

(A) Kidney, (B) spleen, (C) liver, (D) gastrointestinal tract , (E) stomach and (F) heart. All organs were harvested at the age of P18. Millimeter rulers are shown as scale reference.

Figure S3: Histopathological analyses of *Ant2* hypomorphic mice

(A) Gastrointestinal tract, (B) heart, (C) liver, (D) Kidney, (E) spleen and (F) bone were harvested from P1-21 WT or *Ant2*$^{SIBN/Y}$ mice and fixed in formaldehyde. Bone was further decalcified in 10% EDTA (pH 7.5) for 10 days. After embedding in paraffin, the sections were stained with hematoxylin and eosin. Bars represent 100 µm (D; right), (E), (A; stomach), 500 µm (D; left), (A; small intestine and large intestine) and 1mm (F). RV; right ventricle, LV; left ventricle, * indicates muscular ventricular septal defect (VSD).

Figure S4. Cell death analysis in erythroid precursors

Representative images of cell death analysis assessed by flow cytometry. Defined population of proerythroblast (PEB), basophilic erythroblast (BEB), chromatophilic erythroblast (CEB), and orthochromatophilic erythroblast (OCEB) population based on Ter119 and CD71 expression were further stained with AnnexinV and 7-AAD.

Figure S5. Cell death analysis in B cell precursors

Representative images of cell death analysis assessed by flow cytometry. Pro-B cell, pre-B cell and immature B cell populations in bone marrow distinguished by CD43,B220 and IgM expression; and pre-B cell, immature B cell and mature B cell populations in spleen defined by CD43, B220, IgM and IgD expression were stained with 7AAD and Annexin V.

Figure S6: T cell development and death analyses in thymocytes

(A) Thymocytes in WT and *Ant2*$^{SIBN/Y}$ were stained with CD4 and CD8 antibodies and the population was analyzed by flow cytometry. (B) Cell death of total thymocytes were assessed by 7AAD and Annexin V staining.

Figure S7: Phenotype of Ant2 knockout mouse.

Ant2$^{1f/Y}$ embryo at E15.5 (right end) showed pale phenotype as seen in Ant2$^{SIBN/Y}$. Two other Ant2$^{1f/Y}$ embryos in the middle had already died in the uterus prior to sacrifice.